

**Surveillance report on Avian Influenza (AI) conducted in rift valley lakes of Ethiopia**

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## 1. INTRODUCTION

Active disease surveillance is one of the main activities that NAHDIC conducts every year, for diseases which have trans-boundary nature, public health and economic importance that related to livestock export trades. Avian influenza (AI) is a one of trans-boundary disease that potential causes significant impact on animal and human health (zoonosis) and threatens agricultural productivity, food security, and the livelihoods of farming communities in developing countries(FAO, 2011).

Wild birds play important roles in the circulation of avian influenza viruses and are reservoirs for low pathogenic strains. In general, avian influenza viruses in wild birds can be transmitted to and from poultry, and potentially to and from other domestic animals and people (OIE and FAO, 2016).

Ethiopia has a diversified ecology, wet-land and lakes which located in rift valley basin which host several migratory birds in different seasons (FAO 2006) so that pathogenic strain of avian influenza (AI) virus is expected to emerge through migratory birds from other areas.

The current active surveillance program was part of the ongoing Avian influenza (AI) & Newcastle disease (NCD) plan which usually conducted twice a year on risk areas of the country. However, special emphasis was given to this surveillance program due to the current pandemic occurrences of the AI disease (H5N8 subtype) in European, Asian and African countries particularly in neighboring country like Uganda which have put the livelihood of small rural poultry establishments in to a greater risk which historically had been threatened primarily by Newcastle disease viruses. To accomplish this surveillance program, a team composed from molecular biology and Epidemiology was made to carry out the surveillance having with the following objectives.

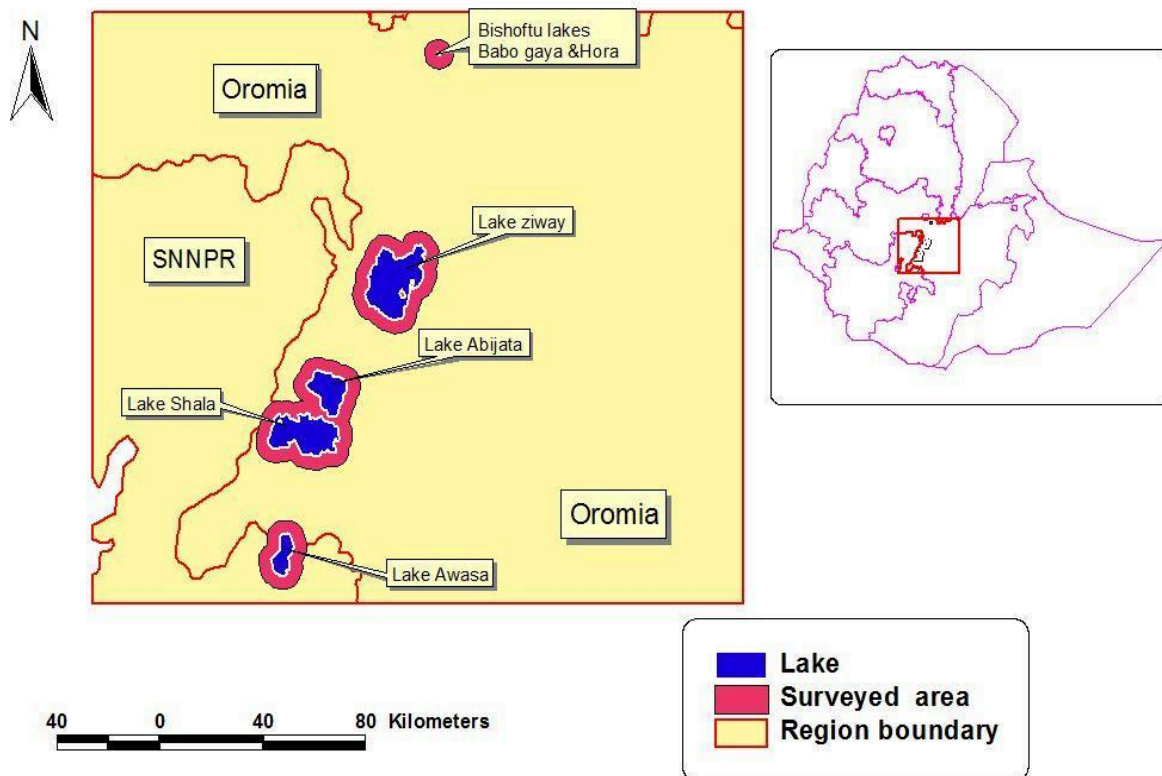
- ✓ To assess the current situation of the disease with in and around the risk areas of the rift valley wet-land areas.
- ✓ For Early detection of the virus strains so as to facilitate rapid and effective control measures aimed at eliminating the virus from the poultry industry

- ✓ To gain a more understanding on the current epidemiology and ecology of avian influenza viruses in risk areas of the country.
- ✓ To create awareness for local poultry farm owners and communities who live around lakes on about the current pandemic occurrences of AI diseases in every corner of the world.

## **2. MATERIALS & METHODS**

### **2.1. Surveillance areas**

The present surveillance program on Avian influenza disease was conducted in lakes located along the rift valley areas where migratory birds are expected to stay temporary. Hora and Babugaya (Bishoftu), Zewaylake (Zeway),Abyata and Shala lakes from Oromia region and Tukurweha & Hawassa lakes from SNNPR region were assessed. Lakes were selected based on the history where different water birds from multiple sources/areas were concentrated. In those assessed areas/lakes, fresh fecal droplets from water birds, swab and serum from poultry farms which have epidemiological linkage to rift valley lakes were collected plus an interview was introduced to fishermen and park security workers who have lived in the areas for a long period of time. In addition, an interview with farm owners was made and at the same time awareness on AI disease was created among poultry farm owners and nearby community.



A map which shows the surveillance area/lakes

## 2.2. Study population

Surveillance was carried out in wild birds and domestic poultry which are more susceptible for avian influenza infections. Animals included in this targeted surveillance were apparently healthy live birds, sick birds, dead birds, and freshly-expelled bird faeces and chickens. Lakes along the rift valley which are previously known as risk areas for the introduction and spread of AI diseases, and poultry farms which have epidemiological linkage to rift valley lakes were targeted for this surveillance program. However, tracheal and cloacal swabs were not collected from water birds due to a reason that birds were not caught/trapped and the team did not get dead birds across the surveillance areas.

### 2.3. Type of surveillance

Targeted surveillance approach on risk areas (lakes) was applied for greater detection efficiency of the disease and cost effectiveness to be achieved. Since severe clinical manifestations, mortality and morbidity are usually observed during infections of highly pathogenic avian influenza infections, **clinical surveillance** was applied for detecting infections of HPAI viruses (OIE and FAO, 2011). However, similar clinical signs, high morbidity and mortality may also be caused by a number of other infectious and non-infectious diseases, **Etiological surveillance** (diagnostic testing at laboratory) was also essentially used for confirming avian influenza infection. In addition low pathogenic avian influenza infection is more difficult to detect through clinical surveillance (unless co-infection is present), etiologically surveillance mainly dependent on diagnostic screening tests with molecular and virological follow up on positive cases was applied (OIE and FAO, 2016).

### 2.4. Sampling methodology and Sample size

Up on the implementation of the targeted surveillance approach, lake Hora, Babugaya, Zeway, shala, Abeyata Tikurweha and Hawassa were the selected lakes where migratory water birds are resting and breeding. In addition it is also expected that these lakes are might be linked through migratory routes to other areas (African, European & Asian) where AI occurrence is currently prevalent. Hence, a total of 153 pool of 5 ( $n=5*153=765$ ) freshly expelled bird feces (265 from Bishuftu, 195 from Zeway, 205 from Abeyata & shalla and 100 from Awasa) were collected. However, tracheal and cloacal swabs were not collected due to the fact that live water birds were not caught/trapped and there were no dead birds across the surveillance areas.

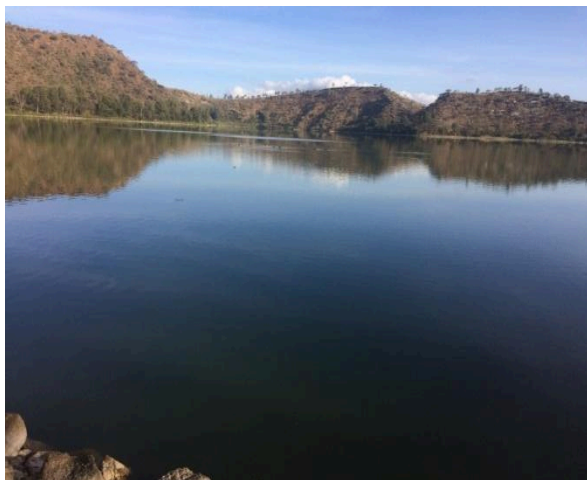
Similarly, risk based sampling was used in domestic poultry farms and markets taking into consideration their location and proximity to the water bodies (lake) which might increase the risk of transmission of avian influenza viruses from migratory water birds to domestic poultry farms. Accordingly, a total of 80 sera and 160 cloacal and tracheal swabs were also collected. Samples were collected in separate cryovial tubes containing viral transport media (VTM). However, Swabs were collected in pairs (Tracheal and cloacal swab pools) from the same

chicken. The specimens were transported to the National Animal Health Diagnostic and Investigation Center (NAHDIC) laboratory and stayed stored at  $-80^{\circ}\text{C}$  until processing.

During sample collection, basic epidemiological information such as location (Latitude and Longitude of each surveillance area, date of sample collection, proximity of lakes to urban poultry/ back yard poultry, community awareness about the diseases, recent wild bird movement, history of morbidity, mortality and clinical signs of birds/chickens, if there is, relevant co-occurrence of the disease in other species including domestic animals and humans were included. However, Species type, sex and age were not possible to include during sampling from water birds.

### 3. FIELD OBSERVATIONS

On the first surveillance day, the team visited Hora and Babugaya lakes (Bishiftu)) and observed a relatively low number of water birds with limited variety/species difference. In addition, the water birds-domestic animals and humans interface was appreciated to be separate. The team did not come across recently dead water birds plus any clinical signs. Water birds were apparently healthy. Additional information was gathered up on an interview of lake security persons with regard to any recent changes on these lakes concerning sickness, death and new movement of water birds. All the interviewee responded that they did not observe anything new on water birds except their number becomes recently decreased which it might be migrated to other lakes.





### Pictures from Hora and Babogaya lakes

However in Zewaylake, aquatic birds population and species variety is observed to be high. The water birds-human-domestic animal interface was highly intermixed (no separation in between). Fishing activities are highly practiced. What the team observed was during evisceration of the trapped fish, the fishery men throw parts of the viscera near around where water birds can easily get and access as their feed. This is the main reason why water birds usually come very close and intermingled with human and domestic animals in the lake.



### Pictures from Zeway lake

Unlike to zeway lake, Abeyata and shala lakes located among the national parks. The team observed when tourists were visiting these lakes. The number of water birds especially in Abayata is considerably high, Flamingo species (European type of bird) were the dominant water birds. There is no fishing activity practice in these lakes. Hence, water birds- human interface is unmixed or separate. Those lakes are relatively faraway (more than 6 km) distance from the backyard domestic poultry which reasonably can be considered as physical barrier for low transmission of AI in case there an incidence of the disease. However, there is a close relation with domestic animals like cattle, goat, sheep and equines as a result of people living in a vicinity practice some small scale farming, grazing land and watering for their live-stocks. During sample collection, the team observed no any sick or recently dead wild birds.



### Pictures from Shala and Abayatalakes

In Awassa Lake especially at Tikurweha and along the sides, there is a similar situation with lake zeway, there is highly diversified and populated wild birds available, fisheries either in individual or in groups were greatly practiced. The team observed while feeding fish byproduct expelled by fishery men from the small meal hoses from the vicinity of the lake. In addition, certain population of wild birds spent (sleep) at night on large trees found at the center of Awassa city.



Pictures from Tikurwuha (Hawassa)

In general the water bird-human-domestic animal interface is not separate. Since wild birds are known as reservoirs and possibly can maintain an avian influenza virus infection, hence close ecological and environmental interaction between wild and domestic is significantly epidemiologically important for the transmission and rapid spread of the disease whenever AI outbreaks may happen.

#### **4. LABORATORY TEST FINDINGS**

Based on the standard operating procedures of NAHDIC, a total of 1005 samples (765 fresh fecal droplets, 160 cloacal and tracheal swabs, 80 sera) were submitted to sample reception unit of NAHDIC, registered and given unique lab code and then submitted to molecular biology Diagnosing lab. Accordingly, two types of test were conducted to detect a circulating virus strain of AI and Newcastle disease from wild birds and backyard chickens.

##### **4.1. Tests for previous infection**



To know the evidence of previous AI infection, sera were tested for sub-type antibody to the H or N antigens using a haemagglutination inhibition (HI) test. Accordingly, all sera samples were found negative for avian influenza virus antibody.

#### **4.2. Molecular test analysis (PCR)**

All fecal droplets and swab samples were analyzed by Real-time Polymerase Chain Reaction (RT-PCR) for detection of avian influenza viral nucleic acid. In addition, swab samples were also tested with real time PCR for Newcastle Diseases virus after extraction of RNA. The result of real time PCR test revealed that all feces droplet and swab samples were negative for avian influenza virus nucleic acid. However, from swab samples, 10 were found positive for Newcastle disease virus nucleic acid.

#### **5. CONCLUSION AND THE WAY FORWARD**

During the surveillance program carried out from 20 to 30 February, 2017, seven lakes which are situated along the rift valley line were surveyed, about 765 fresh expelled droplets from water bird, 160 cloacal and tracheal swabs and 80 sera from poultry farms and markets which have geographical linkage with lakes (water birds) were collected. All samples were processed in molecular biology lab either to detect the evidence of previous infection (serum) or to detect the viral nucleic acid (droplets & swabs). However, the test results of all fresh faecal droplet and swab samples revealed negative for avian influenza virus nucleic acid and also sera samples negative for avian influenza Antibody. Hence, the result of this present surveillance showed that the most risk areas in Ethiopia are currently free of highly pathogenic avian influenza virus. This implies avian influenza disease is not introduced to Ethiopia.

Based on the test result and field observations, the team forwarded the following recommendations.

- ✓ Community need to be aware on immediate reporting of notifiable diseases like HPAI to nearby animal health offices if cases similar with AI are observed in their farms/areas.

- ✓ In an event of massive wild bird death happen, local poultry farms should be advised to implement appropriate bio-security measures.
- ✓ All relevant findings from surveillance have to be shared with relevant stakeholders (animal health, wildlife and public health) authorities at the appropriate time.
- ✓ The task force groups with other relevant stakeholders need to be in a position to adequately and timely respond for appropriate action whenever there is such threaten situations occurring globally.
- ✓ Joint risk communication strategy (among veterinary, wildlife and public health authorities) need to be developed for rapid detection of avian influenza so that a rapid and coordinated response can be implemented upon finding AI new cases.

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